

Excerpt From Diversity of Marine and Freshwater Algal Toxins

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Paralytic Shellfish Toxins

Paralytic shellfish poisoning (PSP) is the most widespread algal derived shellfish poisoning on a worldwide basis (Figure 1). The toxins responsible for PSP are a suite of heterocyclic guanidines collectively called saxitoxins (Figure 2), of which there are currently over 21 known congeners. The crystal structure of the parent compound, saxitoxin, was first described by Schantz et al. (3). The structures of saxitoxin congeners vary by differing combinations of hydroxyl and sulfate substitutions at four sites on the molecule (R1-4). Based on substitutions at R4, the saxitoxins can be subdivided into four groups, the carbamate toxins, sulfo-carbamoyl toxins, and decarbamoyl, and deoxydecarbamoyl toxins. Substitutions at R4 result in substantial changes in toxicity, with the carbamate toxins being the most potent (892-2483 mouse units (MU)/mmol), the decarbamoyl toxins being intermediate in potency (1274-1872 MU/mmol), and the sulfo-carbamoyl toxins generally being the least potent (15-239 MU/mmol) (4).

Paralytic shellfish toxins are produced by a number of genera of gonyaulacoid or gymnodinioid dinoflagellates, including *Alexandrium*, *Gymnodinium*, and *Pyrodinium*, and have also been found in freshwater cyanobacteria (considered later in this chapter). Several studies suggest that saxitoxins can be produced autonomously by bacteria isolated from cultures of PSP producing dinoflagellates (5,6,7), although chemical identity of the toxic activity present in these bacteria has not been unambiguously confirmed. In addition, endosymbiotic or cell-associated bacteria may play a role in the production of paralytic shellfish toxins by dinoflagellates (for review, see 8). Paralytic shellfish toxins are produced in varying proportions by different dinoflagellate species, and even by different isolates within a species. All congeners are not found in any one species. Metabolic pathways responsible for STX biosynthesis have been identified by radio tracer studies (4), which indicate that positions 1-12 of the STX molecule (Figure 2) are formed by the condensation of acetate and arginine. The most prevalent congeners in dinoflagellates, on a molar basis, are the sulfocarbamoyl derivatives. This is of particular interest because the occurrence of N-sulfated groups is rare among natural products (9), and may represent another novel dinoflagellate biosynthetic mechanism. N-sulfotransferase and N-oxidase activities have been identified in STX-producing dinoflagellates (10).

The toxin composition in bivalve tissues can differ markedly from that found in the dinoflagellates ingested. The capacity to metabolize PSP toxins varies substantially between shellfish species (for review, see 11). In most shellfish, PSP toxins with a hydroxysulfate at the C11 position undergo epimerization from the β -epimers (GTX3,4 and C2,4) to the α -epimers (GTX1,2 and C1,2) (9). Of public health significance, the N-sulfocarbamoyl derivatives, which account for the majority of toxin in some dinoflagellates, may be metabolically converted to the more potent decarbamoyl congeners when metabolized in some shellfish species.

The role toxicity plays in the life history of the dinoflagellate species which produce them is not clear. It has been suggested that saxitoxins may play a role in nitrogen metabolism, since some strains accumulate toxin up to 60 pg/cell, or about 0.2% of total wet weight. However, the occurrence of healthy non-toxic strains would suggest that the saxitoxins are secondary metabolites which are not essential for dinoflagellate growth (12).

Saxitoxin binds with high affinity ($K_d \sim 2$ nM) to site 1 on the voltage dependent sodium channel, inhibiting channel opening. The binding affinity of saxitoxin congeners to site 1 varies proportionally with their toxicity in mice (13). The voltage dependent sodium channel plays a critical role in neurotransmission at both the neuronal synapses and neuromuscular junctions. The polarity of the STX molecule largely excludes it from traversing the blood brain barrier; therefore, the primary site of STX action in humans is most likely at the neuromuscular junction. This is consistent with the rapid onset (less than one hour) of symptoms which are classical for paralytic shellfish poisoning, including: tingling and numbness of the perioral area and extremities, loss of motor control, drowsiness, incoherence, and in the case of high doses, respiratory paralysis. The lethal dose in humans is 1-4 mg STX equivalents (14). Clinical symptoms of PSP in humans occurs when approximately 2000 MU (0.72 mg) are ingested, and serious cases generally involve ingestion of 5000 – 20,000 MU toxin (0.9-3.6 mg) (15). In a study of clinical samples from a PSP outbreak in Alaska in 1994 (16), the clearance of saxitoxins from the blood in humans was found to be less than 24 hours, even in patients who had experienced respiratory paralysis and were maintained on life support. Clearance was largely via the urine.

There is currently no widely available antidote for PSP. Anti-STX monoclonal antibodies tested in vitro and in situ show some protection against STX binding and STX-induced reduction of peripheral nerve action potential in rat tibial nerve, suggesting that antibodies may potentially provide useful reagents for protection against toxicity in vivo (17,18). In addition, the potassium channel blocker, 4-aminopurine, has recently been shown to significantly reverse the effects of STX-toxicosis in rats, suggesting that it may be useful as an antidote for PSP (19).

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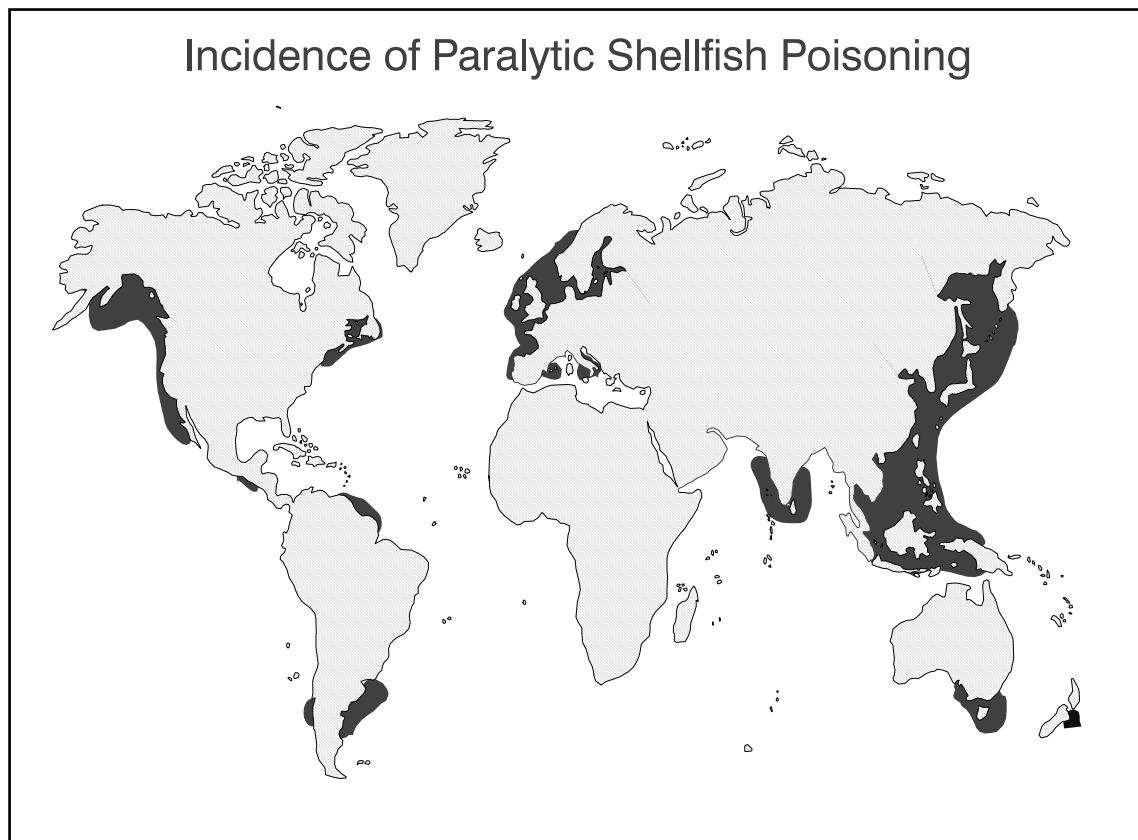
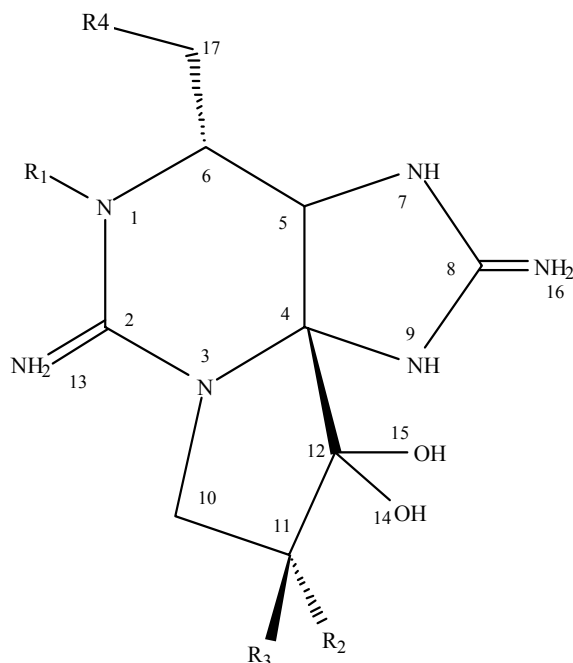


Figure 1. Worldwide incidence of paralytic shellfish poisoning.



		R1	R2	R3	R4	MU/ μmol^1
Carbamate	STX	H	H	H	OCONH ₂	2483
	Neo STX	OH	H	H	OCONH ₂	2295
	GTX1	OH	OSO ₃ -	H	OCONH ₂	2468
	GTX2	H	OSO ₃ -	H	OCONH ₂	892
	GTX3	H	H	OSO ₃ -	OCONH ₂	1584
	GTX4	OH	H	OSO ₃ -	OCONH ₂	1803
Sulfocarbamoyl	GTX5 (B1)	H	H	H	OCONHSO ₃ -	160
	GTX6 (B2)	OH	H	H	OCONHSO ₃ -	-
	C1	H	OSO ₃ -	H	OCONHSO ₃ -	15
	C2	H	H	OSO ₃ -	OCONHSO ₃ -	239
	C3	OH	OSO ₃ -	H	OCONHSO ₃ -	33
	C4	OH	H	OSO ₃ -	OCONHSO ₃ -	143
Decarbamoyl	dcSTX	H	H	H	OH	1274
	dcNeoSTX	OH	H	H	OH	-
	dcGTX1	OH	OSO ₃ -	H	OH	-
	dcGTX2	H	OSO ₃ -	H	OH	1617
	dcGTX3	H	H	OSO ₃ -	OH	1872
	dcGTX4	OH	H	OSO ₃ -	OH	-
Deoxydecarbamoyl	doSTX	H	H	H	H	-
	doGTX2	H	H	OSO ₃ -	H	-
	doGTX3	H	OSO ₃ -	H	H	-

¹Oshima, 1995

Figure 2. Structures and relative toxicity of the paralytic shellfish toxins. Toxicity values are in mouse units (MU), where 1 mouse unit is the amount of toxin required to kill a 20 g mouse in 15 min.